invention is also directed to methods for so improving plant growth by, inter alia, introducing a nucleic acid molecule encoding the bacterial asparagine synthetase into the plant genome (e.g. into plant cells and culturing and/or regenerating the cells into the plants) wherein the nucleic acid molecule is operably linked to a nucleic acid molecule comprising regulatory sequences for expression and for import of the bacterial asparagine synthetase into the chloroplast and/or plastid; and, to plants, having such improved growth.

Claim 12 has been rejected under 35 U.S.C. § 112, second paragraph, as indefinite due to its recitation of "plant, seeds, **and** propagation material".

In response, Applicants have amended claim 12 to more clearly define Applicants' subject matter. The amended wording is supported by the specification at page 7, lines 9-10, which recites that "... seeds, propagule or propagation material of the foregoing methods are contained in the foregoing cells". The "foregoing methods" as well as the "foregoing cells" refer to the transgenic plant material exhibiting the genetic modification of interest. Therefore, Applicants' recitation of "propagation material" means the transgenic plant material, and does not - and indeed cannot - refer to any non-living material, for example, agar and the like.

Applicants submit that the instant amendment overcomes the rejection under 35 U.S.C. §112, second paragraph, which rejection should now be reconsidered and withdrawn.

Claims 9-16 have been rejected under 35 U.S.C. §103(a) as unpatentable over Coruzzi et al., in view of Dudits et al, Temple et al., and Della-Cioppa. The Examiner maintains that Coruzzi et al. suggest producing plants which express a transgene encoding asparagine synthetase and an antisense construct which inhibits production of glutamine synthetase (p. 22, lines 16-19). Coruzzi et al. is said to suggest that nitrogen assimilation in plants could be

improved by targeting asparagine synthetase to chloroplasts (p. 22, lines 3-5), and that it suggests expressing microbial enzymes (p.23, lines 2-7). It is admitted in the Action, however, that Coruzzi et al. does not disclose a chloroplast transit peptide sequence, and that does it not disclose working examples of the transgenic plants.

Dudits et al. is said to disclose transgenic plants expressing a prokaryotic asparagine synthetase. The plants are said to exhibit increased growth relative to normal plants, and the increased growth is even more pronounced in the presence of glutamine synthesis inhibitors.

Temple et al. is cited as teaching that plants contain several forms of glutamine synthetase, and that leaf tissue contains predominantly the chloroplast form of glutamine synthetase (p. 317, col. 2). Temple et al. is said to have successfully reduced glutamine synthetase activity in leaf tissue by expression of an antisense construct. Della-Cioppa et al. is said to teach that a number of chloroplast transit peptide sequences were known in the art (pp. 965-966).

The Examiner therefore maintains that it would have been obvious to one of ordinary skill in the art to have produced plants expressing a prokaryotic asparagine synthetase with a chloroplast transit peptide and an antisense construct to inhibit glutamine synthetase expression.

The Examiner argues that there would have been a reasonable expectation of success, given the demonstrated ability of prokaryotic asparagine synthetase to increase plant growth (Dudits et al.) and the successful inhibition of glutamine synthetase activity by antisense expression (Temple et al.). It is mentioned in the Action that would have been obvious to use

chloroplast transit peptides disclosed by Della-Cioppa et al. to target the asparagine synthetase to chloroplasts, and that the invention as a whole is allege *prima facie* obvious.

In response, Applicants maintain that the rejection under 35 U.S. C. §103(a) is unwarranted and should be reconsidered and withdrawn.

A reference must be considered for all it discloses, including teachings which diverge and teach away from the invention. *In re Dow Chem. Co.*, 5 USPQ2d 1529 (Fed. Cir. 1988). It is impermissible within the framework of § 103 to pick and choose from any one reference only so much as it will support a given position to the exclusion of other parts necessary to the full appreciation of what such a reference fairly suggests to one of ordinary skill in the art. *In re Hedges*, 228 USPQ 685 (Fed. Cir. 1986). In the present situation, it is submitted that none of the references cited, either alone or in any fair combination, serve to obviate the instant invention. Indeed as discussed more fully below, it is urged that a proper reading of the references would lead one of skill away from the instant invention.

Specifically, Applicants respectfully point out that the teachings of Coruzzi, (see page 22, line 16-19 concerning the antisense construct which inhibits production of glutamine synthetase), must be read in connection with the previous sentence, i.e., "in plant species that encode multiple GS isozymes, this may require the suppression of endogenous genes". This clearly reflects the fact that "the endogenous GS genes" which means "all GS genes" must be suppressed. Furthermore, it is a fact that multigene families exist in plants in case of enzymes exhibiting an enzymatic activity of a glutamine synthetase (GS), and it is further a fact, that there are several kinds of GS (i.e., at least GS_1 (cytosolic) and GS_2 (plastidic) present in plants and active in different tissues (see Temple et al. Mol. Gen. Genet. (1993), 236, 315-325).

Applicants have explained in the application (see examples 4 and 5 of the instant specification) that the only GS to be inhibited is chloroplastic GS (GS_2), because only the chloroplastic GS-coding sequence was cloned in the antisense direction.

This object is completely different from Coruzzi's intent to inhibit all GS genes. Consequently, the teachings of Coruzzi et al. differ totally from present invention as there is a completely different overall effect if one inhibits only a part of a set of genes or if one inhibits all genes. Therefore, it is not obvious for a skilled artisan to reduce the knockout-recommendation of Coruzzi (knockout of all GS genes in a plant) to the selected knockout of only a certain species of GS genes (i.e., the chloroplastic GS genes) as taught in present invention.

With respect to Temple et al., Applicants submit that Temple, either alone or together with Coruzzi et al., or with any of the other cited references, actually demonstrates that one of skill did **not** understand how to control selectively the GS_1 and GS_2 genes via an antisense approach. Temple was not able to selectively knockout only GS_1 (see Summary: "Leaves of the plants transformed with the antisense GS_1 construct showed a significant decrease in the level of both GS_1 and GS_2 polypeptides and activity but did not show any significant decrease in the level of endogenous GS mRNA.").

Furthermore, it is stated in Temple at page 319, right column, to page 320, last paragraph:

"Our results suggest that the antisense alfalfa GS transcript does not lower the steady-state level of the endogenous GS transcript in the heterologous plant, tobacco. Since a significant decrease in GS [*i.e. GS_1 and GS_2*] proteins is observed in the GS_1 antisense plants, it is likely that inhibition occurs at the level of translation".

Additionally, see the following statement at page 322, 2d paragraph of Temple et al.:

"The results presented in this paper demonstrate that the full-length alfalfa GS_1 gene, when transcribed in an antisense orientation from the 35S promoter, is capable of down regulating both GS_1 and GS_2 in tobacco leaves. Besides demonstrating that the antisense RNA approach is effective in silencing GS gene expression, our results also show that a heterologous antisense GS_1 transcript is effective in inhibiting both GS_1 and GS_2 gene expression."

These statements demonstrate that while Temple et al. may have been able to inhibit all GS emzymatic activities in a transgenic plant, they were not able to selectively inhibit only one type of GS gene, as taught in present invention.

In contrast to the teachings of Temple et al., Applicants use a homologous gene in an antisense orientation (for example GS_2 from maize to downregulate the transcription of a GS_2 in maize (see example 5) -- they do not use a heterologous approach as Coruzzi et al. describes by using an alfalfa antisense transcript in tobacco plants.

Consequently, Temple et al. - even in combination with Coruzzi et al. - would leave the skilled artisan with the assumption that one would not be able to conduct such a highly selective control of specific GS activities; this is contrary to what is taught in the instant invention. Therefore, it is submitted that Temple et al. either alone or in connection with any of the other cited references, fails to obviate the instant invention.

Dudits' use of at least one chemical compound to inhibit GS-activity, likewise not selective as is the GS_2-antisense construct taught by Applicants. This is because the chemical(s) described by Dudits et al. inhibit(s) GS_1 as well as GS_2 and, therefore, do not exhibit the highly specific inhibition of the inventive GS_2-antisense construct. Consequently, Dudits et al., either

alone or in combination with any of the other cited references, fails to obviate the instant invention.

Although Della-Cioppa et al. discusses several possible chloroplast transit peptides, their efficiency differs significantly from that of the various chimeric proteins (see page 965 right column, 4th paragraph, to page 966, left column, end of 2d paragraph. It is extremely important to combine the optimum transit peptide with the protein of interest to be transferred into the chloroplast, as described by Applicants in "Example 1" of present invention. Consequently, Applicants submit there was no hint, or suggestion in Della-Cioppa either alone or together with any of the other cited references, of the instant invention. The inventive chimeric protein was not described by Della-Cioppa et al., and it was not expected that, for example, this new combination of a transit peptide with the ASN-A obtained from E. coli would be imported into the choloroplast in an efficient way.

Applicants respectfully request therefore that the Section 103 rejection be reconsidered with the following in mind. First, it is well established that "there must be some reason for the combination other than the hindsight gleaned from the invention itself". Uniroyal v. Rudkin-Wiley, 5 U.S.P.Q. 2d 1434, 1438 (Fed. Cir. 1988). Second, there must be some prior art teaching which would have provided the necessary incentive or motivation for modifying the primary reference in the manner suggested by the Examiner. In re Laskowski, 12 U.S.P.Q. 2d 1397, 1399 (Fed. Cir. 1989). Third, "obvious to try" is not the standard under 35 U.S.C. §103. In re Fine, 5 U.S.P.Q. 2d 1596, 1599 (Fed. Cir. 1988). Further, as stated by the Court In re Fritch, 23 U.S.P.Q. 2d 1780, 1783-1784 (Fed. Cir. 1992):

> The mere fact that the prior art may be modified in the manner suggested by the Examiner does not make the modification obvious unless the prior art suggests the desirability of the modification.

The references relied upon by the Examiner fail to provide the necessary incentive or motivation for modifying the references in a manner which would produce the invention as claimed.

In view of the foregoing, Applicants respectfully urge that the rejection does not establish a *prima facie* case of obviousness.

Moreover, even if a prima facie case of obviousness were established (although Applicants do *not* admit this for the reasons set forth above), Applicants have provided evidence of surprising and unobvious results (see instant specification, at inter alia, Example 8, "Growth behavior of transgenic maize and tobacco plants").

It is thus asserted that the claimed invention and its unexpected and surprising advantages are not taught or suggested by the cited references, either individually or in any fair combination. Accordingly, reconsideration and withdrawal of the rejection under 35 U.S.C. § 103(a) are respectfully requested.

Applicants take this opportunity to thank the Examiner for acknowledging Applicants' claim for priority under 35 USC Section 119 and for acknowledging receipt of the priority documents received from the International Bureau.

Please charge any additional fees required or credit any overpayment to Deposit Account No. 50-0320.

Respectfully submitted,

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